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COST IN U.S. DOLLARS
                                                             ENTRY
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FULL ESTIMATED COST
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E2
              1 --> E.C. 4.1.2.11/CN
E3
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E7
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1 E.C. 4.1.2.17/CN
1 E.C. 4.1.2.18/CN
1 E.C. 4.1.2.19/CN
1 E.C. 4.1.2.2/CN
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E9
E10
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=> s e3

1 "E.C. 4.1.2.11"/CN L1

=> d

E11 E12

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ANSWER 1 OF 1 REGISTRY COPYRIGHT 2003 ACS
L1
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9075-38-1 REGISTRY RN

Lyase, hydroxymandelonitrile (9CI) (CA INDEX NAME) CN

OTHER NAMES:

(S)-p-Hydroxymandelonitrile lyase CN

E.C. 4.1.2.11 CN

Hydroxynitrile lyase CN

S-Hydroxynitrilase CN

S-Hydroxynitrile lyase CN

CNs-Oxynitrilase

MF Unspecified

MAN CI

STN Files: AGRICOLA, BIOBUSINESS, BIOSIS, CA, CAPLUS, CASREACT, CEN, LC CHEMINFORMRX, CIN, PROMT, TOXCENTER, USPATFULL

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***

68 REFERENCES IN FILE CA (1957 TO DATE)

2 REFERENCES TO NON-SPECIFIC DERIVATIVES IN FILE CA

68 REFERENCES IN FILE CAPLUS (1957 TO DATE)

=> set expand cont SET COMMAND COMPLETED

=> sel l1 name

E13 THROUGH E18 ASSIGNED

=> index bioscience
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COST IN U.S. DOLLARS

SINCE FILE TOTAL ENTRY SESSION 6.99 7.20

FULL ESTIMATED COST

INDEX 'ADISCTI, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, AQUASCI, BIOBUSINESS, BIOCOMMERCE, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CANCERLIT, CAPLUS, CEABA-VTB, CEN, CIN, CONFSCI, CROPB, CROPU, DDFB, DDFU, DGENE, DRUGB, DRUGLAUNCH, DRUGMONOG2, ...' ENTERED AT 09:59:31 ON 08 MAY 2003

66 FILES IN THE FILE LIST IN STNINDEX

Enter SET DETAIL ON to see search term postings or to view search error messages that display as 0* with SET DETAIL OFF.

=> s e13-18 and immobil?

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- 1 FILE BIOBUSINESS
- FILE BIOSIS
- 9 FILES SEARCHED...
 - 9 FILE BIOTECHABS
 - 9 FILE BIOTECHDS
 - 4 FILE BIOTECHNO
 - 2 FILE CABA
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F6	5	BIOSIS
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=> file f2-20 COST IN U.S. DOLLARS

SINCE FILE TOTAL ENTRY SESSION 15.40 22.60

FULL ESTIMATED COST

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FILE 'TOXCENTER' ENTERED AT 10:16:01 ON 08 MAY 2003 COPYRIGHT (C) 2003 ACS

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- 4 FILES SEARCHED...
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58 L3 L4

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26 DUP REM L4 (32 DUPLICATES REMOVED)

ANSWERS '1-12' FROM FILE CAPLUS

ANSWERS '13-17' FROM FILE IFIPAT ANSWERS '18-22' FROM FILE BIOTECHDS

ANSWER '23' FROM FILE SCISEARCH

ANSWERS '24-25' FROM FILE CEN

ANSWER '26' FROM FILE WPIDS

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DUPLICATE 1 ANSWER 1 OF 26 CAPLUS COPYRIGHT 2003 ACS L5

1998:533933 CAPLUS AN

129:272310 DN

Biocatalysis in microstructured lyotropic liquid crystals ΤI

Boy, M.; Voss, H. ΑU

Institute of Biotechnology, SFB Biocatalysis, Graz University of CS Technology, Graz, A-8010, Austria

Journal of Molecular Catalysis B: Enzymatic (***1998***), 5(1-4), SO 355-359

CODEN: JMCEF8; ISSN: 1381-1177

- Elsevier Science B.V. PB
- Journal DT

English LA Biphasic liq. crystal systems consisting of org. solvent, water and AB surfactant are interesting media for biocatalysis in a non-aq. environment. The application of such systems for the (***S***)catalyzed synthesis of ***lyase*** ***hydroxynitrile*** (S)-mandelonitrile is demonstrated. Screening a favorable liq. crystal system is the first step. Exptl. of the influence of temp., enzyme and substrate concn. on the kinetics are presented. Interactions of the matrix and the mass ***immobilization*** three-dimensional liq. cryst. transfer and biochem. reaction kinetics are shown. THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS RECORD RE.CNT 7 ALL CITATIONS AVAILABLE IN THE RE FORMAT DUPLICATE 2 ANSWER 2 OF 26 CAPLUS COPYRIGHT 2003 ACS L5 1998:691672 CAPLUS AN130:135818 DN ***lvase*** ***hydroxynitrile*** Expression of the Zn2+-containing TI from flax (Linum usitatissimum) in Pichia pastoris- utilization of the recombinant enzyme for enzymic analysis and site-directed mutagenesis Trummler, Klaus; Roos, Jurgen; Schwaneberg, Ulrich; Effenberger, Franz; ΑU Forster, Siegfried; Pfizenmaier, Klaus; Wajant, Harald Institute of Cell Biology and Immunology, University of Stuttgart, CS Stuttgart, 70569, Germany Plant Science (Shannon, Ireland) (***1998***), 139(1), 19-27 SO CODEN: PLSCE4; ISSN: 0168-9452 Elsevier Science Ireland Ltd. PΒ Journal DTEnglish LA (HNL) are involved in the ***lyases*** ***Hydroxynitrile*** AB catabolism of cyanogenic glycosides in cyanogenic plants and are powerful tools in the stereoselective synthesis of cyanohydrins. The recent ***lyase*** from flax (Linum cloning of the ***hydroxynitrile*** usitatissimum; LuHNL) reveals that this enzyme defines a novel class of HNL. Thorough biochem. and mutational anal. of LuHNL have been hampered by low expression levels of the recombinant enzyme in Escherichia coli. To overcome this impediment, we have cloned a myc-His-tagged LuHNL-cDNA under control of the methanol-inducible AOX1 (alc. oxidase) promoter of Pichia pastoris and introduced it in the SMD1168 strain. Recombinant LuHNL was kinetically indistinguishable from the authentic flax enzyme. ***Immobilized*** LuHNL was used for synthesis of several aliph. (R)-cyanohydrins in a preparative scale to analyze the products according to enantiomeric excess and yield of reaction. LuHNL has significant homologies to members of the Zn2+-contg. alc. dehydrogenases (Zn2+-ADHs). In particular, residues responsible for coordination of Zn2+ ions or fulfilling structural or functional tasks in Zn2+-ADHs are conserved. found about 2-4 mol zinc per mol of recombinant LuHNL using atom absorption spectroscopy in a non His-tagged version of LuHNL. Using site-directed mutagenesis, we substituted several of the conserved residues against alanine in LuHNL and found that in most cases, HNL-activity was impaired. Hence, it seems that LuHNL and Zn2+-ADHs have similar structural requirements with respect to maintaining a catalytically active structure. Residues essentially involved in catalysis of Zn2+-ADHs are also of functional importance in LuHNL, suggesting that the removal of the proton from alc. and cleavage of cyanohydrins can be fulfilled by similar active site structures. THERE ARE 23 CITED REFERENCES AVAILABLE FOR THIS RECORD RE.CNT 23 ALL CITATIONS AVAILABLE IN THE RE FORMAT

1997:591161 CAPLUS AN127:175496 DN Production of (S)-cyanohydrins TI Effenberger, Franz; Wajant, Harald; Foerster, Siegfried; Roos, Juergen IN Degussa Ag, Germany PΑ Ger. Offen., 11 pp. SO CODEN: GWXXBX \mathbf{DT} Patent German LΑ FAN.CNT 1 APPLICATION NO. DATE KIND DATE PATENT NO. ______ ______ DE 1997-19703314 19970130 <--A1 19970814 DE 19703314 PΙ EP 1997-101401 19970130 <--A2 19971008 EP 799894 A3 19991208 EP 799894 R: AT, BE, CH, DE, DK, FR, GB, LI, NL JP 09227488 A2 19970902 JP 1997-23666 19970206 <--US 1997-796873 19970207 <--A 19990323 US 5885809 19960209 PRAI DE 1996-19604715 MARPAT 127:175496 OS (S)-Cyanohydrins are produced in isomeric excess from the corresponding AΒ carbonyl compds. by reaction with HCN or a CN- salt catalyzed by (***S***) - ***oxynitrilase*** (EC 4.1.2.37) ***immobilized*** nitrocellulose. DUPLICATE 4 ANSWER 4 OF 26 CAPLUS COPYRIGHT 2003 ACS L5 1995:1004311 CAPLUS AN 124:80260 DN Purification and characterization of a novel (R)-mandelonitrile lyase from ΤI the fern Phlebodium aureum Wajant, Harald; Foerster, Siegfried; Selmar, Dirk; Effenberger, Franz; ΑU Pfizenmaier, Klaus Institut fuer Zellbiologie Immunologie, Universitaet Stuttgart, Stuttgart, CS 70569, Germany Plant Physiology (***1995***), 109(4), 1231-38 SO CODEN: PLPHAY; ISSN: 0032-0889 American Society of Plant Physiologists PΒ Journal DTLA Using high-performance liq. chromatog. and NMR we identified vicianin as AB the cryogenic compd. of Phlebodium aureum. The (R) - ***hydroxynitrile*** ***lyase*** involved during cyanogenesis in the catabolism of the aglycon ([R]-mandelonitrile) was purified to apparent homogeneity. The purified holoenzyme is a homomultimer with subunits of Mr = 20,000. At least three isoforms of the enzyme exist. In contrast to other ***lyases*** , mandelonitrile lyase (MDL) from ***hydroxynitrile*** P. aureum was not inhibited by sulfhydryl- or hydroxyl-modifying reagents, suggesting a different catalytic mechanism. The enzyme is active over a broad temp. range, with max. activity between 35 and 50.degree. and a pH optimum at 6.5. In contrast to (R)-MDLs isolated from several species of the Rosaceae family, (R)-MDL from P. aureum is not a flavoprotein. The substrate specificity was investigated using ***immobilized*** enzyme and diisopropyl ether as solvent. The addn. of cyanide to arom. and heterocyclic carbonyls is catalyzed by this (R)-MDL, whereas aliph. carbonyls are poorly converted.

ANSWER 3 OF 26 CAPLUS COPYRIGHT 2003 ACS

L5

DUPLICATE 3

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1993:254558 CAPLUS
ΑN
DN
    118:254558
    Process for preparing optically active cyanohydrins with enzymes
ΤI
    Andruski, Stephen W.; Goldberg, Bruce
IN
    FMC Corp., USA
\Delta
    U.S., 7 pp.
SO
     CODEN: USXXAM
\mathbf{DT}
     Patent
     English
LΑ
FAN.CNT 1
                                        APPLICATION NO. DATE
                   KIND DATE
     PATENT NO.
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                                        US 1991-809803
                    A 19930105
                                                          19911217 <--
     US 5177242
PΙ
                                        CA 1992-2093826 19921113 <--
                     AA 19930618
     CA 2093826
                     C
                           19950829
     CA 2093826
WO 9312072
                     A1 19930624 WO 1992-US9945 19921113 <--
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         RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, SE, BF,
             BJ, CF, CG, CI, CM, GA, GN, ML, MR, SN, TD, TG
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     AU 9331414
                      B2 19940331
     AU 647982
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                     T2 19931104
     JP 05507736
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                     A1 19940406
     EP 590096
     EP 590096 B1 19960306
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CZ 1993-651 19921113 <--
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PL 1992-300043 19921113 <--
RU 1993-33481 19921113 <--
RO 1993-489 19921113 <--
CN 1992-114106 19921202 <--
NO 1993-1316 19930406 <--
     HU 71308
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     CZ 280422
                     E 19960315
     AT 135047
                     B1 19961231
     PL 170490
                  C1 19971010
B1 19971230
     RU 2092558
                     B1 19971230
     RO 112765
                     A 19930811
     CN 1075166
                     A 19930816
     NO 9301316
     KR 9709155 B1 19970607
                                        KR 1993-71172 19930420 <--
PRAI US 1991-809803 A
WO 1992-US9945 A
                           19911217
                           19921113
     CASREACT 118:254558; MARPAT 118:254558
OS
     Title compds. esp. (S)-cyanohydrins, useful as intermediates in prepn. of
AB
     known pyrethroid insecticides, are prepd. by an improved process
     comprising reacting aldehydes with HCN in presence of the catalytic enzyme
       ***S*** - ***oxynitrilase*** chem. bound to a porous membrane.
       3-(PhO)C6H4CHO and HCN were reacted at 6.degree. to give the title compds.
      (S)-3-(PhO)C6H4CH(NC)OH and its R-isomer is in a ratio of 96/4. The above
     process does not only increase the selectivity of S- to R-isomers, but
      also shorten the reaction time.
     ANSWER 6 OF 26 CAPLUS COPYRIGHT 2003 ACS
                                                      DUPLICATE 6
 L5
      1992:189955 CAPLUS
 AN
 DN
      116:189955
     Purification and protein characterization of ***hydroxynitrile***
 тT
      ***lyases*** from sorghum and almond
      Jansen, I.; Woker, R.; Kula, M. R.
 ŪΆ
```

ANSWER 5 OF 26 CAPLUS COPYRIGHT 2003 ACS

L5

DUPLICATE 5

Inst. Enzymtechnol., Heinrich-Heine-Univ. Duesseldorf, Juelich, D-5170, CS Biotechnology and Applied Biochemistry (***1992***), 15(1), 90-9 SO CODEN: BABIEC; ISSN: 0885-4513 Journal DT English LΑ ***Hydroxynitrile*** ***lyase*** of Sorghum bicolor (EC 4.1.2.11) AΒ was purified by a new procedure using (NH4)2SO4 pptn. and ***immobilized*** metal ion affinity chromatog. as key steps. The protein was characterized according to mol. wt., subunit compn., pI, pH, and temp. stability and was compared with ***hydroxynitrile*** ***lyase*** of almond (EC 4.1.2.10). Furthermore, the N-terminal sequences of the oxynitrilases are reported. ANSWER 7 OF 26 CAPLUS COPYRIGHT 2003 ACS DUPLICATE 7 L5 1993:228768 CAPLUS ΑN 118:228768 DN Purification of ***S*** - ***oxynitrilase*** from Sorghum bicolor by TΙ ***immobilized*** metal ion affinity chromatography on different carrier materials Woker, R.; Champluvier, B.; Kula, M. R. ΑU Inst. Enzymetechnol., Heinrich-Heine-Univ. Duesseldorf, Juelich, D-5170, CS Journal of Chromatography, Biomedical Applications (***1992***), SO 584(1), 85-92 CODEN: JCBADL; ISSN: 0378-4347 Journal DTEnglish LA The purifn. of ***hydroxynitrile*** ***lyase*** (EC 4.1.2.11, AB ***S*** - ***oxynitrilase***) from S. bicolor is compared using different strategies. A new procedure is presented, which exploits the affinity of ***S*** - ***oxynitrilase*** towards metal ions as a key step in purifn. The metal ions are ***immobilized*** by chelators on different carrier materials, e.g. Sepharose beads, microporous membranes or poly(ethylene glycol). A systematic examn. demonstrates the excellent potential of ***immobilized*** metal affinity chromatog. as a preparative sepn. method. ANSWER 8 OF 26 CAPLUS COPYRIGHT 2003 ACS DUPLICATE 8 L_5 1990:215459 CAPLUS $\mathbf{A}\mathbf{N}$ 112:215459 DN One carbon homologation of carbohydrates by two-phase TI transcyanohydrination Arena, Blaise J. IN Allied-Signal, Inc., USA PΑ U.S., 9 pp. Cont.-in-part of U.S. Ser. No. 810,627, abandoned. SO CODEN: USXXAM DTPatent English LΑ FAN.CNT 1 PATENT NO. KIND DATE DATE APPLICATION NO. DATE _____ _ _ _ _ _____ PI US 4900667 A 19900213 PRAI US 1985-810627 19851219 US 1987-73259 19870713 <--A method for manufg. L-sugars for use in artificial sweeteners by homologating monosaccharides via their cyanohydrins uses

transcyanohydrination across a phase boundary from a water-insol. donor cyanohydrin dissolved in a water-immiscible org. solvent to an aq. soln. of a receptor monosaccharide. This heterogeneous transcyanohydrination can be incorporated into a cyclic process where the cyanide donor is enzymically regenerated via enzyme catalyzed addn. of HCN to a suitable aldehyde. The latter process can be made continuous or semicontinuous by ***immobilization*** of the enzyme. The donor cyanohydrin

mandelonitrile (I) was prepd. enzymically from NaCNand benzaldehyde. I in chloroform was used to homologate L-arabinose to a mixt. of L-glucocyanohydrin and L-mannocyanohydrin with a conversion rate of 70%. By using 5-fold excess of I and proper agitation, a conversion rate of 90% is ensured. Furthermore, most of the cyanide dissocd. from the mandelonitrile did notenter the aq. phase.

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ANSWER 9 OF 26 CAPLUS COPYRIGHT 2003 ACS
L5
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MΑ 1998:493698 CAPLUS

129:135261 DИ

Enzymic processes for preparing (S)-cyanohydrins ΤI

Kirchner, Gerald; Wirth, Irma; Werenka, Christian; Griengl, Herfried; IN Schmidt, Michael

DSM Chemie Linz G.m.b.H., Austria; Kirchner, Gerald; Wirth, Irma; Werenka, PA Christian; Griengl, Herfried; Schmidt, Michael

SO PCT Int. Appl., 38 pp. CODEN: PIXXD2

DTPatent

LA German

FAN.																		
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ΡI	WO	9830	711		A:	1	1998	0716		W	19:	97-E	P2692	2	1997	0526	<	
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	US	6337	196		В	1	2002	0108		U	5 19:	99-3	3176	1	1999	0625		
PRAI	ΑT	1997	-41		Α		1997	0113										
	WO	1997	-EP2	692	W		1997	0526										
ΩS	MΛD	יייעם	129.	1352	5 1													

MARPAT 129:135261 os

The invention concerns an enantioselective process for prepg. the AB (S)-enantiomer of an optically active cyanohydrin by reacting an aldehyde or ketone with a cyanide group donor. According to this process, the aldehyde or ketone is reacted with a cyanide group donor in an org. diluent in the presence of a recombinant (***S***)-

hydroxynitrile ***lyase*** from Hevea brasiliensis, the resultant (S)-cyanohydrin being isolated from the reaction mixt.

THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD

ALL CITATIONS AVAILABLE IN THE RE FORMAT

```
ANSWER 10 OF 26 CAPLUS COPYRIGHT 2003 ACS
L5
    1996:154053 CAPLUS
AN
    124:224570
DN
    The first recombinant ***hydroxynitrile***
                                                  ***lyase***
                                                                 and its
    application in the synthesis of (S)-cyanohydrins
    Foerster, Siegfried; Roos, Juergen; Effenberger, Franz; Wajant, Harald;
ΑU
    Sprauer, Achim
    Inst. Org. Chemie Univ., Stuttgart, D-70569, Germany
CS
    Angewandte Chemie, International Edition in English ( ***1996*** ),
SO
    35(4), 437-9
    CODEN: ACIEAY; ISSN: 0570-0833
    VCH
PB
DT
    Journal
LA
    English
    The authors overexpressed Manihot esculenta ***hydroxynitrile***
AB
      ***lyase*** (meHNL) in Escherichia coli. Enantioselective addn. of
    hydrocyanic acid to several aldehydes and ketones was by enzyme
      ***immobilized*** on nitrocellulose and using diisopropyl ether as
     solvent was demonstrated.
    ANSWER 11 OF 26 CAPLUS COPYRIGHT 2003 ACS
L5
    1993:470564 CAPLUS
AN
DN
    119:70564
    Manufacture of chiral cyanohydrins by resolution with oxynitrilase
ΤI
    Niedermayer, Uwe
IN
PA
    Germany
SO
    Ger., 4 pp.
    CODEN: GWXXAW
DT
    Patent
LA
    German
FAN.CNT 1
                                       APPLICATION NO. DATE
     PATENT NO.
                   KIND DATE
     _____
                                         _____
                                                         ------
                                       DE 1991-4139987 19911204 <--
    DE 4139987
                    C1 19930415
PΙ
                                        WO 1992-DE1018 19921203 <--
                    A1 19930610
     WO 9311255
        W: AU, CA, JP, US
        RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE
                     A1 19930628 AU 1993-40288 19921203 <--
     AU 9340288
                                        EP 1992-924554 19921203 <--
     EP 576638
                     A1 19940105
        R: AT, BE, CH, DE, ES, FR, GB, LI, NL
PRAI DE 1991-4139987 19911204
     WO 1992-DE1018
                          19921203
     Chiral cyanohydrins are prepd. in high yield and enantiomeric purity by
AB
     digestion of racemic mixts. of cyanohydrins with (R) - or ( ***S*** )-
       ***oxynitrilase*** . The HCN produced in this reaction is removed from
     the equil. mixt. by a carbonyl-contg. compd. such as formaldehyde or
     acetaldehyde, thereby promoting the reaction. Racemic mandelonitrile and
     acetaldehyde were incubated with ***immobilized***
                                                         (R)-oxynitrilase.
     (S)-madelonitrile of ee 93.8% was produced in 75% yield.
L5
     ANSWER 12 OF 26 CAPLUS COPYRIGHT 2003 ACS
     1976:86148 CAPLUS
AΝ
DN
     84:86148
ΤI
    Binding of biologically active proteins to carriers
```

Jaworek, Dieter; Maier, Josef; Nelboeck-Hochstetter, Michael

PA Boehringer Mannheim G.m.b.H., Fed. Rep. Ger.

SO Ger. Offen., 25 pp.

CODEN: GWXXBX

DT Patent

LA German

FAN.CNT 1

FAN.CN'	r 1				_
PA	ATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI DI	E 2426988	A1	19760108	DE 1974-2426988	19740604 <
DI	E 2426988	C2	19850214		
A.	Г 7503391	Α	19760915	AT 1975-3391	19750502 <
A:	Г 336530	В	19770510		
GI	3 1450519	Α	19760922	GB 1975-22659	19750523 <
US	5 4038140	Α	19770726	US 1975-582495	19750530 <
CI	H 621146	Α	19810115	CH 1975-7038	19750530 <
N	L 7506507	Α	19751208	NL 1975-6507	19750602 <
Fl	R 2273816	A1	19760102	FR 1975-17461	19750604 <
Fl	R 2273816	В1	19790323		
J:	P 51012891	A2	19760131	JP 1975-67421	19750604 <
PRAI D	E 1974-2426988		19740604		

AB Enzymes were bound covalently to carriers by carrying out copolymn. of activated polysaccharide and hydrophilic monomer or copolymn. of a monomer mixt. (without polysaccharide) in the presence of enzyme. One of the monomers polymd. with polysaccharide was reacted with the enzyme directly and thus the enzyme would be covalently bound to carrier. Alternatively, a crosslinking agent coupled to the enzyme was used to link enzyme with a grafted copolymer (e.g. starch-acrylamide). Thus, acrylamide and starch allyl ether were dissolved in phosphate buffer. Then, a soln. of D-

hydroxynitrile ***lyase*** (from almonds) with which acrylic acid chloride had been reacted was added to the starch-acrylamide buffered soln. Next, the polymn. starter soln. contg. 5% ammonium peroxydisulfate and 5% 3-dimethylaminopropionitrile was added. After the polymerizate set for 3 hr, it was pressed through a sieve of mesh size 0.4 mm and eluted into a column with buffered 0.5M NaCl. Other enzymes bound by similar procedures were yeast hexokinase, swine pancreatic trypsin, and acylase I of swine kidney.

- L5 ANSWER 18 OF 26 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI
- AN 1997-03392 BIOTECHDS
- TI Enantioselective synthesis of aliphatic (S)-cyanohydrins in organic solvents using ***hydroxynitrile*** ***lyase*** from Manihot esculenta;

cassava recombinant enzyme purification from Escherichia coli (conference paper)

- AU Wajant H; Forster S; Sprauer A; Effenberger F; Pfizenmaier K
- CS Univ.Stuttgart-Inst.Cell-Biol.Immunol.; Univ.Stuttgart-

Inst.Org.Chem.Isotop.Res.

- LO Institut fuer Zellbiologie und Immunologie, Universitaet Stuttgart, 70569 Stuttgart, Germany.
- SO Ann.N.Y.Acad.Sci.; (***1996***) 799, 771-76

CODEN: ANYAA9 ISSN: 0077-8923

Enzyme Engineering XIII, San Diego, CA, 15-20 October, 1995.

- DT Journal
- LA English
- AN 1997-03392 BIOTECHDS
- The purification and expression cloning of acetone-cyanohydrin-lyase from cassava (Manihot esculenta Crantz) (MeHNL, EC-4.1.2.37) is described.

The MeHNL gene was cloned in the expression vector plasmid pQE3 to give plasmid pQE3-MeHNLwt, which was used to transform Escherichia coli M15(pREP4) cells for overexpression of MeHNL. Expression of MeHNL was induced by adding IPTG. Recombinant MeHNL was purified using Q-Sepharose column, gel filtration chromatography and anion-exchange chromatography. The specific activity of purified MeHNL was 86 U/mg, and a yield of 0.5 g soluble MeHNL (from an 80 l culture) was achieved (150 mg pure enzyme). The enzyme was purified 5.1-fold, with a yield of 54% and a specific activity of 86 U/mg. The enzyme was used for (S)-cyanohydrin production in disopropyl ether, using HCN as the cyanide source. Use of a highly concentrated MeHNL preparation for ***immobilization*** increased activity 4- to 10-fold. (7 ref)

L5 ANSWER 19 OF 26 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI

AN 1994-13130 BIOTECHDS

TI The potential of lyases in organic synthesis;

e.g. aspartate-ammonia-lyase, L-aspartate-4-decarboxylase, fumarase and phenylalanine-ammonia-lyase application in the chemical and food industry (conference paper)

AU van den Tweel W J J; van der Werf M J; Hartmans S; Schoemaker H E; Kamphuis J; de Bont J A M

CS DSM-Res.; Univ.Wageningen-Agr.

- LO Bio-organic Chemistry Section, DSM Research, P.O. Box 18, 6160 MD Geleen, The Netherlands.
- SO Prog.Biotechnol.; (***1994***) 9, Pt.1, 455-62 CODEN: PBITE3

DT Journal

- LA English
- AN 1994-13130 BIOTECHDS
- The following conversions using lyases were discussed: (1) production of AΒ L-aspartic acid from fumaric acid using ***immobilized*** Escherichia coli aspartate-ammonia-lyase (EC-4.3.1.1); (2) production of L-alanine and D-aspartic acid from L-aspartic acid using ***immobilized*** Pseudomonas dacunhae L-aspartate-4-decarboxylase in a packed column reactor; (3) production of L-malic acid from ***immobilized*** Brevibacterium flavum fumarate-hydratase (EC-4.2.1.2); (4) production of L-phenylalanine using Rhodotorula rubra phenylalanine-ammonia-lyase (EC-4.3.1.5); and (5) beta-hydroxylation of carboxylic acids. Examples of achievements in research and development include: (i) production of R-***S*** - ***oxynitrilase*** ; (ii) and S-cyanohydrins using R- and acyloin formation mediated by benzoylformate-decarboxylase (EC-4.1.1.7); (iii) production of D-malic acid and D-citramalic acid from maleic acid and citraconic acid, respectively using Pseudomonas pseudoalcaligenes NCIMB 9867 malease; and (iv) carbon-carbon formation by transketolases (EC-2.2.1.1). (19 ref)
- L5 ANSWER 20 OF 26 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI

AN 1994-01125 BIOTECHDS

- TI Enzyme-catalyzed C-C linkage using oxynitrilases and aldolases;

 (R) and (***S***) ***oxynitrilase*** , fructose-1,6diphosphate-aldolase and 3-hexulose-6-phosphate-synthase (conference paper)
- AU Kula M R; Albrecht J; Beisswenger R; Brockamp H P; Jansen I; Niedermeyer U

CS Univ.Heinrich-Heine-Duesseldorf.Inst.Enzymtechnol.

LO Heinrich-Heine-Universitaet Duesseldorf, Institut fuer Enzymtechnologie, Postfach 2050, D-5170 Juelich, Germany.

DECHEMA Monographies; (***1993***) 185-95 SO CODEN: 9999W DTJournal German LΑ 1994-01125 BIOTECHDS ΑN Oxynitrilases from 3 plant sources were purified to give final activities AB of 69-260 U/mg. During conversion of carbonyl compounds with HCN, the enzymes from almond (Prunus amygdalus) husk and flax (Linum usitatissimum) gave (R)-cyanohydrins whilst that from Sorghum bicolor yielded (S) enantiomers. Racemic products were also obtained under many reaction conditions, but if the pH was suitably adjusted, optical purity was significantly increased. (R) - and (***S***)on lyotropic liquid crystals ***immobilized*** ***oxynitrilases*** gave 88% purity at pH 7 and 99% and pH 4.5. ***Immobilized*** (R)-oxynitrilase in a continuous fixed bed reactor produced D-mandelonitrile with 94-96% ee over 100 hr. The (S)-enzyme converted mostly benzaldehyde derivatives and the (R)-oxynitrilases had wider specificities. A fructose-1,6-diphosphate-aldolase (EC-4.1.2.13) isolated from Staphylococcus carnosus is a class I aldolase with a broad specificity for aldehydes as substrates and with good stability. 3-Hexulose-6-phosphate-synthase from Methylomonas sp. M 15 had stereoselectivity for aldol condensation with unnatural substrates including the production of octuloses. (32 ref) ANSWER 21 OF 26 BIOTECHDS COPYRIGHT 2003 THOMSON DERWENT AND ISI 1.5 1992-09334 BIOTECHDS AN Enzymatic syntheses of optically active (R) - and (S) -cyanohydrins in ΤI organic solvents; stereospecific cyanohydrin production in an organic phase system using ***immobilized*** almond mandelonitrile-lyase or Sorghum bicolor hydroxymandelonitrile-lyase (conference paper) Effenberger F; Foerster S; Hoersch B; Ziegler T ΑU Institute of Organic Chemistry, University, Pfaffenwaldring 55, D-W 7000 LO Stuttgart 80, Germany. Biochem.Eng.Stuttgart; (***1991***) 134-36 SO Journal DT English LA 1992-09334 BIOTECHDS AN (R)-oxynitrilase (mandelonitrile-lyase, EC-4.1.2.10) from defatted bitter AB almond flour and (***S***) - ***oxynitrilase*** (hydroxymandelonitrile-lyase, EC-4.1.2.11) from Sorghum bicolor seedlings were used to catalyze asymmetric (R) - and (S) -cyanohydrin production, respectively, in an organic phase system. The enzymes were on Avicel cellulose. (S)-cyanohydrins were produced ***immobilized*** (***S***)- ***oxynitrilase*** ***immobilized*** by suspension of in 10 ml diisopropyl ether, addition of aldehyde (2 mmol) and HCN (300 ul, 7.5 mmol), and stirring until all of the aldehyde had reacted. With (R)-oxynitrilase, (R)-cyanohydrins of aromatic, aliphatic and heterocyclic aldehydes were obtained in 96-99% ee and excellent yield (71-90%). (***S***)- ***oxynitrilase*** showed a greater substrate specificity, accepting exclusively aromatic aldehydes as substrates, and had a slower reaction rate. (S)-cyanohydrins were

1-amino-2-alcohols and alpha-hydroxyaldehydes. (10 ref)

obtained in 89-97% ee and 80-93% yield. (R)-and (S)-cyanohydrins are useful intermediates for synthesis of alpha-hydroxycarboxylic acids,

AN 1990-05832 BIOTECHDS
TI Enzyme-catalyzed synthesis of (S)-cyanohydrins and subsequent hydrolysis to (S)-alpha-hydroxy-carboxylic acids;

stereospecific cyanohydrin production using ***immobilized***
hydroxymandelonitrile-lyase

AU Effenberger F; Hoersch B; Foerster S; Ziegler T

LO Institut fuer Organische Chemie, Universitaet Stuttgart, Pfaffenwaldring 55, D-7000 Stuttgart 80, Germany.

SO Tetrahedron Lett.; (***1990***) 31, 9, 1249-52 CODEN: TELEAY

DT Journal

LA English

AN 1990-05832 BIOTECHDS

(hydroxymandelonitrile-lyase, AB (***S***)- ***Oxynitrilase*** EC-4.1.2.11) was used to catalyze enantioselective addition of HCN to aldehydes yielding (S)-cyanohydrins in very high optical purity. High optical yields were only obtained if non-enzymatic addition, which resulted in racemic production, was successfully suppressed by working in organic solvents e.g. ethyl acetate or diisopropyl ether. Acid-catalyzed hydrolysis of optically active cyanohydrins afforded alpha-hydroxy carboxylic acids in optically active form. The enzyme-catalyzed reaction for production of (S)-cyanohydrins involved addition of a solution of hydroxymandelonitrile-lyase to a stirred suspension of Avicel-cellulose in 0.05 M phosphate buffer containing (NH4)2SO4. The mixture was stirred enzyme suspended at RT for 10 min, filtered and the ***immobilized*** in diisopropyl ether. After addition of aldehyde and HCN, the mixture was stirred until all of the aldehyde had reacted. The

immobilized enzyme was removed and the filtrate was concentrated to yield (S)-cyanohydrin. In a typical reaction, 3-phenoxymandelonitrile (0.42 g) was obtained from 3-phenoxybenzaldehyde (0.40 g). (21 ref)

L5 ANSWER 23 OF 26 SCISEARCH COPYRIGHT 2003 THOMSON ISI

AN 1999:48339 SCISEARCH

GA The Genuine Article (R) Number: BM15C

TI Oxynitrilases: From cyanogenesis to asymmetric synthesis

AU Schmidt M (Reprint); Griengl H

CS GRAZ TECH UNIV, SPEZIALFORSCHUNGSBEREICH BIOKATALYSE, STREMAYRGASSE 16, A-8010 GRAZ, AUSTRIA (Reprint); GRAZ TECH UNIV, INST ORGAN CHEM, A-8010 GRAZ, AUSTRIA

CYA AUSTRIA

TOPICS IN CURRENT CHEMISTRY, (***OCT 1999***) Vol. 200, pp. 193-226.

Publisher: SPRINGER-VERLAG BERLIN, HEIDELBERGER PLATZ 3, W-1000 BERLIN 33,
GERMANY.

ISSN: 0342-6793.

DT General Review; Journal

FS PHYS

LA English

REC Reference Count: 230

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

Oxynitrilases are enzymes which catalyse the formation and cleavage of cyanohydrins. The cyanohydrin formation reaction proceeds by stereoselective addition of hydrogen cyanide to aldehydes or ketones to give enantiopure alpha-hydroxynitriles. This simple method of C-C bond formation has become a promising method to obtain a number of biologically active compounds. Cyanohydrin fission plays an important role in nature and is involved in plant defence where hydrogen cyanide is liberated upon plant damage. Among the known oxynitrilases only the (R)-oxynitrilase from

Prunus amygdalus and the (***S***)- ***oxynitrilases*** brasiliensis and Manihot esculenta are available in sufficient quantities which allow cyanohydrin formation on a larger scale. Prunus amygdalus oxynitrilase can easily be isolated from natural sources (bitter almond bran) and for two (***S***)- ***oxynitrilases*** functional overexpression allows their production in sufficient amounts for broad preparative applications. The three dimensional structure of the (***S***)- ***oxynitrilase*** from Hevea brasiliensis has been determined, and suggestions concerning the reaction mechanism have been discussed. Several procedures employing oxynitrilases have been developed to date which enable cyanohydrin formation on a preparative scale, particularly the use of buffer solutions as the reaction medium, organic solvents with ***immobilised*** enzymes, as well as biphasic reaction systems. Possible follow up reactions of the generated hydroxy and nitrile functionality, as well as the conversion of unsaturated cyanohydrins into valuable asymmetric compounds are outlined.

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L5 ANSWER 26 OF 26 WPIDS (C) 2003 THOMSON DERWENT
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AN 1991-275467 [38] WPIDS

CR 1991-209314 [29]

DNC C1991-119359

Prepn. of optically active cyanohydrin derivs. - by enzymatic reaction of oxo cpds. with hydrocyanic acid using oxy nitrilase solubilised in liq. crystal.

DC B05 C02 C03 D16 E19

IN MIETHE, P; WANDREY, C; KRAGL, U; KULA, M R; STURTZ, I M; STUERTZ, I M

PA (KERJ) FORSCHUNGSZENT JUELICH GMBH; (UYHA-N) UNIV MARTIN LUTHER HALLE-WITTENBERG; (UYMA-N) MARTIN-LUTHER UNIV; (KERJ) FORSCHUNGSZENTRUM JUELICH GMBH

CYC 11

A 19910918 (199138)* PΙ EP 446826 R: CH DE FR GB IT LI NL SE DE 4008411 A 19910926 (199140) DE 4028689 A 19920312 (199212) 6p <--US 5122462 A 19920616 (199227) 7p <--DE 4008411 C2 19930708 (199327) 5p <--A3 19920715 (199334) EP 446826 <--A 19940215 (199411) JP 06038793 7p <--EP 446826 B1 19951115 (199550) DE 10p <--R: CH DE DK FR GB LI NL DE 59106876 G 19951221 (199605)

ADT EP 446826 A EP 1991-103667 19910311; DE 4008411 A DE 1990-4008411 19900316; DE 4028689 A DE 1990-4028689 19900910; US 5122462 A US 1991-670437 19910318; DE 4008411 C2 DE 1990-4008411 19900316; EP 446826 A3 EP 1991-103667 19910311; JP 06038793 A JP 1991-51393 19910315; EP 446826 B1 EP 1991-103667 19910311; DE 59106876 G DE 1991-506876 19910311, EP 1991-103667 19910311

FDT DE 59106876 G Based on EP 446826

PRAI DE 1990-4028689 19900910; DE 1990-4008411 19900316; DE 1990-4008412 19900316

AN 1991-275467 [38] WPIDS

CR 1991-209314 [29]

AB EP 446826 A UPAB: 19951221
Prepn. of optically active cyanohydrin derivs. (I) comprises enzymatic reaction of oxo cpds. with hydrocyanic acid in an organic solvent in the presence of (R) - or (S) -oxy-nitrilasee (4.1.2.10) or (4.1.2.11) solubilised in a lyotropic liq. crystal. Reaction takes place under

acidic conditions and such that competing chemical reactions and racemisation are excluded. The surfactants used in the liq. crystal formation are not those whose hydrolysis would lead to an increase in pH.

USE/ADVANTAGE - (I) can be used in the prepn. of optically active alpha-aminoalcohols, alpha-hydroxycarboxylic acids, heterocycles and pyrethroid insecticides. (I) are easily derivated chiral building blocks which can be obtd. economically in sufficiently large amounts with the highest possible enantiomer excess(ee). The use of the lyotropic liq. crystal ensures that the reaction can be carried out under favourable conditions (esp. without the need to use low temps.) and with relatively low enzyme loss. @(9pp Dwg.No.0/2)

ABEQ US 5122462 A UPAB: 19930928

Prepn. of optically-active cyanohydrins (I) comprises solubilising (R)-oxynitrilase (4.1.2.10) or (***S***)- ***oxynitrilase*** (4.1.2.11) in a lyotropic liq. crystal, the oxynitrilase catalysing the prodn. of (I). An oxo cpd. is enzymatically reacted with hydrocyanic acid in an organic solvent in the presence of the solubilised oxynitrilase under conditions sufficiently acid for the competing chemical reaction and racemisation to be negligible.

Surfactants which produce an increase in pH upon hydrolysis are excluded from the liq. crystal formation. Pref. the reaction is carried out in a surfactant/organic solvent/liq. buffer system which has been prepd. using buffer solns. with pH values of 3-6.

USE - (I) are useful in the prodn. of optically-active alpha-amino alcohols, alpha-hydroxy carboxylic acids, heterocycles and pyrethroid insecticides.

ABEQ DE 4008411 C UPAB: 19931116

The flow reactor for heterogeneous conversion of fluids contg. substrate using lyotropic liq. crystals has a porous tube, filled with liq. crystals and a sealed end. The tube is contained in a sliding fit within a shrouding tube which forms the reactor mantle. The mantle forms a flow channel for the fluid flow, through a gap together with the pore vol. of the tube. The fluid enters through a feed distributor, and emerges through an upper outlet. the dia. of the inner tube is determined by the diffusion speed of the substrate at the biocatalyst, and is structured so that virtually all the liq. crystal layer is involved in the reaction.

USE/ADVANTAGE - Apps. uses the lyotropic liq. crystals to ***immobilise*** enzymes or microorganisms in organic solns. The assembly gives a high yield for usage time of the reactor vol. with no loss of activity in the reactor filling.

Dwq.0/2

ABEQ EP 446826 B UPAB: 19951215

Process for producing optically active cyanohydrins by enzymatic reaction of oxo compounds with hydrogen cyanide in the presence of (R) - or (

S) - ***oxynitrilase*** (4.1.2.10) and (4.1.2.11) respectively under such acid conditions that the competing chemical reaction and racemisation are negligible, characterisd in that the reaction is carried out in an organic solvent in the presence of oxynitrilase solubilised in a lyotropic liquid crystal, excluding, in respect of the formation of the liquid crystal, those tensides and hydrolysis of which results in an increased pH.

Dwg.0/2

L5 ANSWER 13 OF 26 IFIPAT COPYRIGHT 2003 IFI
3627298 ENZYMATIC PROCESSES FOR PREPARING (S)-CYANOHYDRINS; REACTION OF KETONE
WITH CYANIDE TO FORM CYANOHYDRIN. Griengl Herfried (AT); Kirchner Gerald
(DE); Schmidt Michael (AT); Werenka Christian (AT); Wirth Irma (AT). DSM
Fine Chemicals Austria GmbH AT (49459).
US 6337196 8 Jan 2002.
PCT Pub. No. WO 9830711 16 Jul 1998. APPLICATION: US 1999-331761 25 Jun
1999.
PCT Appl. No. WO 1997-EP2692 26 May 1997. PCT 371 date 25 Jun 1999, PCT
102(e) date 25 Jun 1999.

PRIORITY: AT 1997-41 19970113. TYPE OF PATENT: UTILITY. FILE SEGMENT: CHEMICAL; GRANTED. No. of Claims: 9

L5 ANSWER 14 OF 26 IFIPAT COPYRIGHT 2003 IFI
3127080 METHOD OF PRODUCING (S)-CYANOHYDRINS; CATALYTIC CYANATION IN THE
PRESENCE OF AN ***IMMOBILIZED*** OXYNITRILASE WITH NITROCELLULOSE
CARRIER IN ORGANIC SOLVENT. Effenberger Franz (DE); Forster Siegfried
(DE); Roos Jurgen (DE); Wajant Harald (DE). Degussa DE (23568).
US 5885809 23 Mar 1999.
APPLICATION: US 1997-796873 7 Feb 1997.
PRIORITY: DE 1996-19604715 19960209.
TYPE OF PATENT: UTILITY; REASSIGNED. DOCUMENT TYPE: CERTIFICATE OF
CORRECTION. CORRECTION DATE: 9 Nov 1999. FILE SEGMENT: CHEMICAL; GRANTED.
No. of Claims: 9
MICROFILM REEL-FRAME NOS: 008554-0019.

L5 ANSWER 15 OF 26 IFIPAT COPYRIGHT 2003 IFI
2714660 ENANTIOMERIC ENRICHMENT OF CYANOHYDRINS; SELECTIVE DEHYDROCYANATION
WITH OXYNITRILASE ENZYME. van Eikeren Paul. Bend Research Inc (10802).
US 35230 7 May 1996. US 5241087 31 Aug 1993 ORIGINAL PATENT.
APPLICATION: US 1994-192867 7 Feb 1994. US 1992-848023 9 Mar 1992
ORIGINAL APPLICATION.
TYPE OF PATENT: REISSUE. DOCUMENT TYPE: CERTIFICATE OF CORRECTION.
CORRECTION DATE: 15 May 2001. FILE SEGMENT: CHEMICAL; GRANTED.
No. of Claims: 19 Graphics Info: 5 Drawing Sheet(s), 5 Figure(s).

L5 ANSWER 16 OF 26 IFIPAT COPYRIGHT 2003 IFI
2393723 ENANTIOMERIC ENRICHMENT OF CYANOHYDRINS. van Eikeren Paul. Bend
Research Inc (10802).
US 5241087 31 Aug 1993.
APPLICATION: US 1992-848023 9 Mar 1992.
TYPE OF PATENT: UTILITY; REISSUE REQUESTED. FILE SEGMENT: CHEMICAL;
GRANTED. OTHER SOURCE: CA 120:8336.
No. of Claims: 19 Graphics Info: 5 Drawing Sheet(s), 5 Figure(s).
MICROFILM REEL-FRAME NOS: 006049-0896.

L5 ANSWER 17 OF 26 IFIPAT COPYRIGHT 2003 IFI
2262702 PROCESS FOR THE ENZYMATIC PREPARATION OF OPTICALLY-ACTIVE
CYANOHYDRINS; REACTING OXO COMPOUND WITH HYDROCYANIC ACID IN PRESENCE OF
OXYNITRILASE SOLUBILIZED IN LYOTROPIC LIQUID CRYSTAL UNDER ACID
CONDITIONS. Kragl Udo (DE); Kula Maria-Regina (DE); Miethe Peter (DE);
Stuertz Ingeborg M (DE); Wandrey Christian (DE). Forschungszentrum Julich
GmbH DE (24899).
US 5122462 16 Jun 1992 (CITED IN 004 LATER PATENTS).
APPLICATION: US 1991-670437 18 Mar 1991.
PRIORITY: DE 1990-4008411 19900316; DE 1990-4008412 19900316; DE

1990-4028689 19900910.
TYPE OF PATENT: UTILITY; EXPIRED. FILE SEGMENT: CHEMICAL; GRANTED.
No. of Claims: 8 Graphics Info: 1 Drawing Sheet(s), 2 Figure(s).
MICROFILM REEL-FRAME NOS: 005713-0494.

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